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***Hymenochaete* in China. 8. *H. biformisetosa* sp. nov.
with a key to species with denticulate setae**

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ABSTRACT — *Hymenochaete biformisetosa*, a new species from Yunnan Province in southwestern China, is described and illustrated. It is characterized by thick, resupinate basidiocarps, two types of setae (sometimes denticulate), and ellipsoid to broadly ellipsoid basidiospores. The new species is similar to *H. spinulosestosa* and *H. denticulata* but with morphological and molecular (ITS rDNA sequences) differences. An identification key to nine species of *Hymenochaete* with denticulate setae is provided.

KEY WORDS — *Hymenochaetaceae*, phylogeny, taxonomy, wood-inhabiting fungi

Introduction

The diversity of wood-inhabiting fungi in Yunnan Province, southwest China, is extremely high because of abundant vascular plants and complex landforms. Although many basidiomycete species have been described from this area in recent years (Dai et al. 2002; Yuan & Dai 2008a,b; Dai & Li 2010, Dai 2011, 2012; He & Li 2011, 2012, He & Dai 2012; Zhao et al. 2013), corticioid fungi are under-collected and studied.

In 2012, an intensive investigation of wood-inhabiting fungi was carried out in the Gaoligong Mountains in northwest Yunnan Province, which is considered an important biodiversity hotspot in China. Among the approximately 100 specimens collected in this survey, an undescribed species of *Hymenochaete* was found. We describe and illustrate the new fungus as *H. biformisetosa* in our new contribution to the knowledge of *Hymenochaete* species in China.

Materials & methods

MORPHOLOGICAL STUDIES: Voucher specimens were deposited in the herbarium of Beijing Forestry University (BJFC), and the microscopic procedure followed Dai (2010).

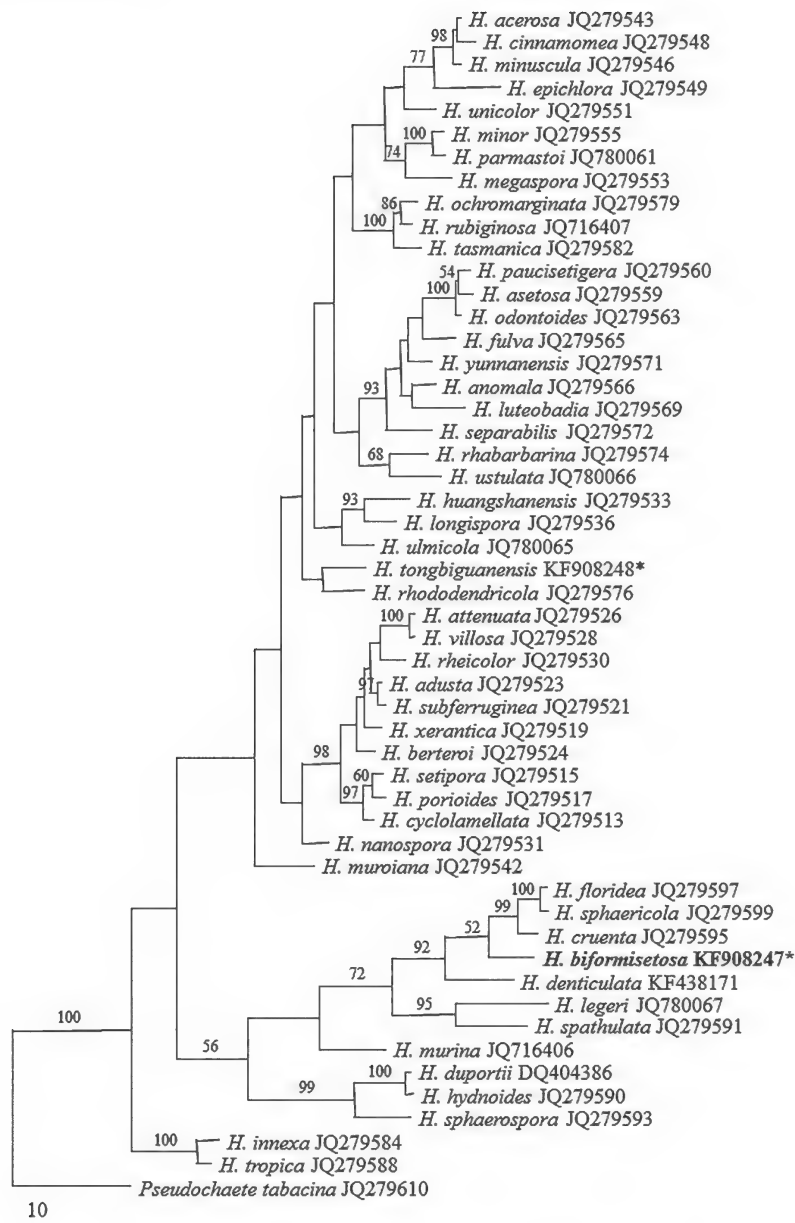


FIG. 1. Strict consensus tree obtained from Maximum Parsimony analysis of ITS sequences of *Hymenochaete* species. *Pseudochaete tabacina* was used as outgroup. Newly generated sequences are indicated by an asterisk (*). Parsimony bootstrap values >50% are shown.

In presenting the size range of spores and setae, 5% of the measurements from each end of the range are given in parentheses. Special color terms follow Petersen (1996).

DNA EXTRACTION AND SEQUENCING: A CTAB Plant genome rapid extraction kit DN14 (Aidlab Biotechnologies Co., Ltd) was used to extract DNA from dried specimens following the manufacturer's instructions. Approximately 700 base pairs of the ITS rDNA were amplified with primers ITS5 and ITS4 as follows: initial denaturation at 95 °C for 3 min, followed by 38 cycles of 94 °C for 40 s, 58 °C for 45 s and 72 °C for 1 min, and a final extension of 72 °C for 10 min. DNA sequencing was performed at Beijing Genomics Institute, and all the newly generated sequences were deposited in GenBank.

PHYLOGENETIC ANALYSIS: The phylogeny was inferred from the ITS sequences of 51 *Hymenochaete* species. *Pseudochaete tabacina* (Sowerby) T. Wagner & M. Fisch. was selected as outgroup. The sequences were aligned using the ClustalX 1.83 (Chenna et al. 2003). Alignments were optimized manually in BioEdit 7.0.5.3 (Hall 1999). Maximum parsimony analysis were performed using PAUP* 4.0b10 (Swofford 2002). Gaps in the alignments were treated as missing data. Trees were generated using 100 replicates of random stepwise addition of sequence and tree-bisection reconnection (TBR) branch-swapping algorithm, with all characters given equal weight. Branch supports for all parsimony analysis were estimated by performing 1000 bootstrap replicates (Felsenstein 1985) with a heuristic search of 10 random-addition replicates for each bootstrap replicate. The tree length (TL), consistency indices (CI), retention indices (RI), rescaled consistency indices (RC) and homoplasy index (HI) were calculated for each tree generated. Trees were figured in Treeview 1.6.6 (Page 1996).

Phylogenetic results

Two new ITS sequences were obtained in this study (GenBank accession numbers KF908247 and KF908248). The alignment of the ITS sequences of 52 taxa resulted in 764 sites with 389 parsimony informative characters. Two equally parsimonious trees were obtained from the analysis with TL = 2180, CI = 0.423, RI = 0.588, RC = 0.249 and HI = 0.577. The strict consensus tree showed that *Hymenochaete biformisetosa* formed a distinct lineage embedded in a clade with *H. denticulata* J.C. Léger & Lanq., *H. cruenta* (Pers.) Donk, *H. floridea* Berk. & Broome, and *H. sphaericola* Lloyd (FIG. 1).

Taxonomy

Hymenochaete biformisetosa Jiao Yang & S.H. He, sp. nov.

FIGS 2–4, 8

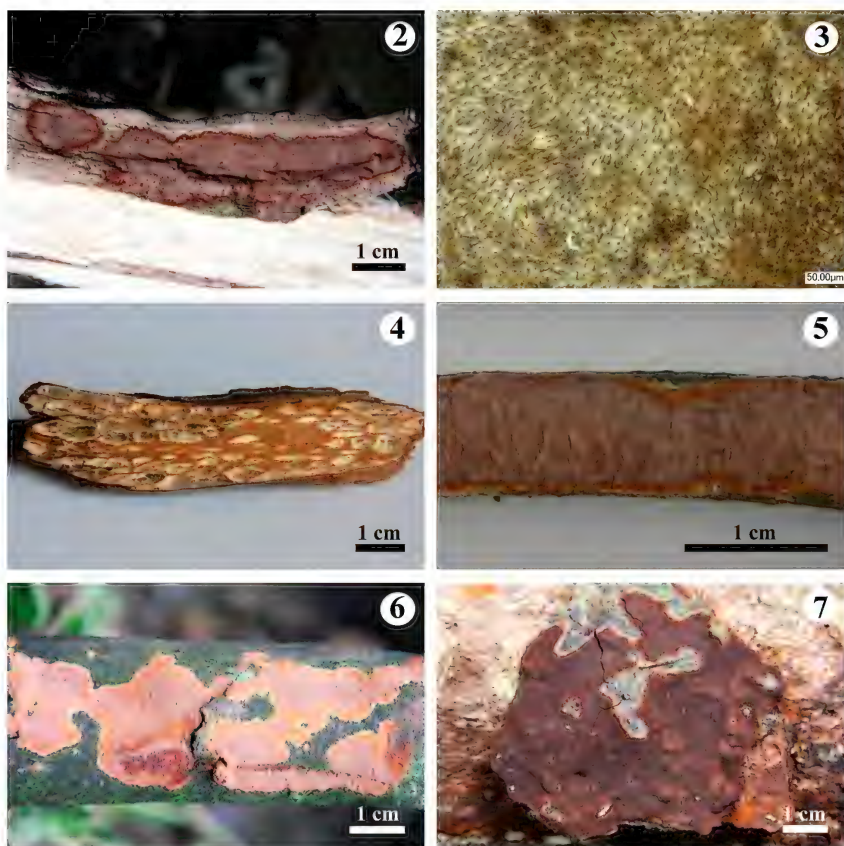
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Differs from other *Hymenochaete* species by its thick basidiocarps, two types of setae that may be denticulate, and ellipsoid to broadly ellipsoid basidiospores.

TYPE: China. Yunnan Prov., Baoshan, Gaoligongshan Nat. Res., Baihualing, on angiosperm stump, 28.X.2012, He 1445 (**holotype**, BJFC 14297; GenBank KF908247).

ETYMOLOGY: the epithet "*biformisetosa*" refers to the two types of setae.

FRUITBODY: Basidiocarps annual, effused, closely adnate, corky to woody hard, first as small round or irregular patches, later confluent up to 6 × 2 cm,



FIGS. 2–3. *Hymenochaete biformisetosa* (holotype): 2. Basidiocarps. 3. Close-up of hymenophore surface. 4. Wood decayed by *H. biformisetosa*. FIGS. 5–7. Basidiocarps of other *Hymenochaete* species with denticulate setae: 5. *H. denticulata* (He 1271). 6. *H. separabilis* (He 271). 7. *H. tongbiguanensis* (He 1552).

0.15–2.5 mm thick in section. Hymenophore smooth, grayish brown to fawn, not cracked when young, becoming pale mouse-gray to light vinaceous gray, deeply and sparsely cracked when mature; margin thinning out, reddish brown to orange-brown, distinct, up to 1 mm wide when juvenile, becoming fuscous or concolorous with hymenophore surface, narrower with age.

HYPHAL STRUCTURE: Hyphal system subdimitic; generative hyphae without clamp connections; tissue darkening but otherwise unchanged in potassium hydroxide.

SUBICULUM: Tomentum and cortex absent. Hyphal layer absent or sometimes present, consist of interwoven hyphae. Generative hyphae rare,

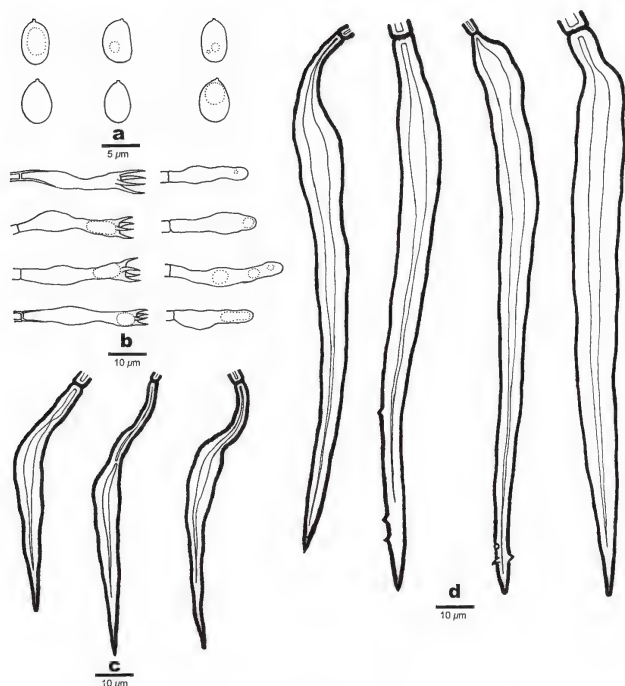


FIG. 8. *Hymenochaete biformisetosa* (holotype):
a. Basidiospores. b. Basidia and basidioles. c. Small setae. d. Large setae.

hyaline to yellowish brown, thin- to slightly thick-walled with a wide lumen, septate, moderately branched, 2–4 µm in diam. Skeletoid hyphae predominant, reddish brown, distinctly thick-walled with a narrow lumen, some parts nearly solid, usually developing into setal hyphae with acute apices, 3–4.5 µm in diam.

STRATIFIED HYMENIUM: Hyphae similar to those in subiculum, reddish brown, thick-walled, agglutinated, interwoven. Setal layer thick, composed of several to many rows of overlapping setae. Setae of two types (1) large, numerous, subulate, reddish brown, with acute tips, projecting up to 90 µm above the hymenium, (70–)80–130(–140) × (8–)9–14(–16) µm, often with crystals loosely attached to the projected part when mature; (2) small, numerous, subulate, reddish brown, with acute tips, some with a hyphal tail, usually not protruding, (30–)35–50(–60) × 4–8 µm. Both types of setae sometimes with 1–3 conical spines near apex; spines up to 2 µm long. Cystidia and hyphidia absent. Basidia subcylindrical to subclavate, usually restricted in central part, with four sterigmata and a simple septum at base, 20–25 × 3–5 µm; basidioles similar to basidia in shape but shorter.

SPORES: Basidiospores ellipsoid to broadly ellipsoid, hyaline, thin-walled, smooth, sometimes containing one guttule, inamyloid and nondextrinoid in Melzer's reagent, acyanophilous in cotton blue, $(4.1\text{--})4.3\text{--}6(-6.5) \times (2.8\text{--})3\text{--}4.2(-4.5) \mu\text{m}$, mean spore length = $5.2 \mu\text{m}$, mean spore width = $3.5 \mu\text{m}$, length/width = 1.5 (60 spores of one specimen were measured).

TYPE OF ROT: Causes a white pocket rot of angiosperm wood.

ADDITIONAL SPECIMENS EXAMINED: *Hymenochaete spinulosestosa*: VENEZUELA. MERIDA PROV., Monte Zerpa, on dead hardwood, 29.I.2001, Ryvarden 43707 (isotype, TAAM 202926).

Hymenochaete denticulata: CHINA. GUANGXI AUTONOMOUS REGION, Longzhou County, Nonggang Nat. Res., on fallen angiosperm twig, 21.VII.2012, He 1271 (BJFC 14122; FIG. 5).

Hymenochaete separabilis: CHINA. ANHUI PROV., Huangshan County, Huangshan Nat. Res., on fallen angiosperm trunk, 21.X.2010, He 460 (BJFC 9938; FIG. 6).

Hymenochaete tongbiguanensis: CHINA. YUNNAN PROV., Ruili, on fallen angiosperm branch (paratype, HMAS 84867); Longchuan County, Tongbiguan Nat. Res., on angiosperm stump, 31.X.2012, He 1497 (BJFC 14352). HAINAN PROV., Lingshui County, Diaoluoshan Nat. Res., on angiosperm stump, 10.XI.2012, He 1552 (BJFC 14407; GenBank KF908248; FIG. 7).

Discussion

Hymenochaete biformisetosa can be easily distinguished from other species of the genus by its thick ($\leq 2.5 \mu\text{m}$) resupinate basidiocarps, two types of setae that sometimes are denticulate, and ellipsoid to broadly ellipsoid basidiospores; it causes a distinct white pocket rot of angiosperm wood.

Two species, *Hymenochaete tongbiguanensis* T.X. Zhou & L.Z. Zhao and *H. spinulosestosa* Parmasto, also have thick basidiocarps and denticulate setae. *Hymenochaete tongbiguanensis* can be distinguished from *H. biformisetosa* by its smaller setae ($22.5\text{--}42 \times 3.5\text{--}8 \mu\text{m}$) and basidiospores ($3\text{--}4 \times 1.5\text{--}2 \mu\text{m}$; Zhang & Dai 2005), and *H. spinulosestosa* has large, broadly ellipsoid basidiospores ($5\text{--}6.2 \times 3.2\text{--}3.7 \mu\text{m}$) but only one type of setae ($50\text{--}75 \times 5\text{--}7.5 \mu\text{m}$; Parmasto 2012).

Six other species of *Hymenochaete*, including *H. denticulata*, also produce denticulate setae, but they differ from *H. biformisetosa* in having thin basidiocarps ($<150 \mu\text{m}$), shorter setae ($40\text{--}60 \mu\text{m}$), and shorter basidiospores ($<5 \mu\text{m}$, Léger 1998; Parmasto 2012). *Hymenochaete unicolor* Berk. & M.A. Curtis is similar to *H. biformisetosa* in basidiocarp morphology and anatomical structure with smaller, smooth setae ($35\text{--}60 \times 5\text{--}9 \mu\text{m}$, Parmasto 2005).

In the phylogenetic tree (FIG. 1), *Hymenochaete biformisetosa* appears to be closely related to *H. denticulata* (bootstrap value = 92%), which also has two types of denticulate setae (Léger 1998; He & Li 2014). The differences between these two species are discussed above. The new species is related also to the

H. cruenta group in the phylogenetic tree (FIG. 1); however, species of this group have red-colored basidiocarps, dendrohyphidia, and cylindrical basidiospores (Léger 1998; Parmasto 2001; Parmasto & Gilbertson 2005).

Of the nine *Hymenochaete* species with denticulate setae now described (Gilbertson & Hemmes 1997; Léger 1998; Zhang & Dai 2005; Parmasto 2012), four are known from China (Zhang & Dai 2005; He & Li 2014).

Key to species of *Hymenochaete* with denticulate setae

1. Basidiocarps <0.15 mm thick2
1. Basidiocarps up to 1 mm or more7
2. Well-developed hyphal layer present3
2. Hyphal layer absent.4
3. Setae 30–40 µm long, with 1–3 teeth *H. separabilis*
3. Setae 40–70 µm long, with 5–10 teeth *H. tomentelloidea*
4. Setae with 1–2 teeth *H. pellicula*
4. Setae with several to many teeth5
5. Setae of two types *H. denticulata*
5. Setae uniform.6
6. Cortex present; spores subglobose, 3.7–4.3 µm wide. *H. harpago*
6. Cortex absent; spores ellipsoid to cylindrical, 1.5–2 µm wide *H. pertenuis*
7. Setae <50 µm long; spores 3–4 × 1.5–2 µm *H. tongbiguanensis*
7. Setae >50 µm long; spores >4 × 3 µm.8
8. Setae of two types, large setae up to 130 µm long. *H. biformisetosa*
8. Setae uniform, up to 75 µm long. *H. spinulosoetosa*

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